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Temperature Dependency of Population Dynamics and Trophic Cascades in a Model Aquatic System

Reed Brodersen
University of Nebraska-Lincoln

TEMPERATURE DEPENDENCY OF POPULATION DYNAMICS AND TROPHIC CASCADES IN A MODEL AQUATIC SYSTEM

By

Reed Brodersen

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Reed Brodersen, B.S.

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Advisor: Dr. John DeLong

Determining how temperature alters direct predation and indirect trophic cascade effects will be crucial in understanding how global climate change affects community structure and stability. While we are starting to comprehend the effects of warming on individual trophic interactions and underlying mechanisms, effects of warming on multilevel food webs are mostly unknown. This study sought to experimentally observe potential effects of temperature on population and community dynamics, and the magnitude of trophic cascades. To reach these objectives, a three-tiered food web (*Scenedesmus*, *Daphnia* and *Hydra*) was designed, maintained, and observed at two different temperatures (17°C and 21°C) for 37 days. Results found that *Daphnia* population dynamics shifted, with the 21°C treatments demonstrating higher density peaks, faster population declines, and an additional population cycle over the duration of the study. *Scenedesmus* populations showed no response to differential *Daphnia* population dynamics. While a strong trophic cascade effect was found, no differences were observed between temperatures. The experimental design may have been overly limited to accurately observe trophic cascade effects, so more research is encouraged. Mechanisms for *Daphnia* population dynamic shifts were hypothesized, including temperature-dependent rates of maturation and mortality, reproductive timing and capacity, and *Scenedesmus* productivity. Consequences of temperature-induced population dynamic shifts were explored, concluding that similar effects in vivo may contribute to uncoupling of predator-prey interactions, and increased community instability.

Introduction and Literature Review

Global climate change is set to be the most significant environmental, social and economic challenge in human history. It is also slated to become the most rapid change in climate in known geologic time, which presents a unique set of challenges for natural biological systems that have traditionally adapted to changing climates over hundreds of thousands of years, not decades.

This study will focus on the effects of warming on one important component of ecosystems: food web dynamics. More specifically, it will attempt to determine the temperature dependency of predator-prey dynamics and trophic cascade effects in an aquatic model system. To best understand the relevance of this study, it is important to understand the role of predation in biological systems, and how alterations may induce system-wide effects.

Effects of Climate Change on Biological Systems

Global climate change (GCC) will affect biological systems in a region specific manner, as global climate change expresses itself in highly variable changes in regional temperatures, which may be more of less significant than the global average. Thus, it can be difficult to make generalized statements as to how GCC will affect biological systems. There are many theories, though, as to what some region specific effects may look like.

One important aspect of GCC is that diurnal temperature ranges have already decreased, because minimum temperatures are increasing almost twice as fast as maximum temperatures (Karl et al. 1991). This has serious ramifications for the timing and duration of freeze-free periods. This has both non-biological effects, like increasing ice and snow melt, and biological effects like increased geographic ranges and relative competitiveness of certain (mainly invasive) species (Peterson et al. 2009). Additionally, changes in precipitation are expected in most parts

of the earth, with potential effects ranging from increased water stress on organisms and communities, to entire ecosystem shifts.

Furthermore, Vasseur et al. argue that the increase in temperature variation caused by GCC, and the increased frequency, severity and duration of temperature extremes, will be even more damaging than increased mean temperatures. While some species may benefit from increased mean temperatures, many will suffer losses of performance. (Vasseur 2014)

Walther et al. proposed that GCC will affect four main areas: (1) the phenology (timing of life cycles) and physiology of organisms, (2) the geographic range and distribution of species, (3) the composition of and interactions within communities, and (4) the structure and dynamics of ecosystems (2002). This study will focus on the third component: interactions within communities. More specifically, it will focus on predation and how it affects community structure.

Key Role Of Predators In Shaping Community Structure

Predation, and consumption in general, is the mechanism by which energy and nutrients travel up the food chain. It embodies complex physiological and behavioral processes, and is influenced by a wide range of abiotic and biotic factors. Predation can play an important role in regulating populations, and maintaining community stability. Though, the nature and magnitude of these effects differ by predator, prey type, community structure, etc. Clarke found that general predators have a stabilizing effect on numerous prey populations because they feed preferentially on those species with the greatest abundance (1962). This, in turn, allows for greater species diversity and potential for complexity, which was first empirically confirmed by Paine in 1966. Predation can also play an interesting role in shaping communities through what are known as

trophic cascades, which help explain the significant role key predators play in shaping biological communities (Shurin et al. 2002).

Trophic Cascades

Trophic cascades are indirect positive effects of top predators on producers, caused by the reduction in populations of intermediate consumers. Trophic cascades have been found to have significant impacts in a variety of terrestrial and aquatic ecosystems (Knight et al. 2005). The best known example can be found in the effects of wolf removal on the defoliation and decimation of plants, via uncontrolled deer population growth in areas such as Yellowstone National Park (Ripple and Beschta 2004). In many systems trophic cascade effects can act to increase stability and overall productivity (Carpenter et al 1985). Increased understanding of these processes have led to their use in ecosystem management practices in conservation and agricultural contexts (Barbosa 1998; Huffaker 2012).

Interaction Strength

Interaction strengths are defined and quantified in a variety of ways. Generally, they are the relative effect of a predator or consumer on the abundance of the resource species, though they may also be quantified by parameters of the functional response model (Wootton and Emmerson 2005). They act to quantify the strength of a predator-prey relationship, which allows for better understanding and predictions of important community properties like overall function or stability (Vallina and Le Quéré 2011). In this study, they will be used to help quantify and explain any potential changes in population dynamics or the magnitude of the trophic cascade.

Functional Response: Theoretical Framework for Modeling Predation

Functional Response Model

To study complex predator-prey interactions, it's important to place these into a measurable framework. In this case, we use what is called a functional response model. A functional response is a curve that relates predator kill rates to prey density. Functional responses are a core ecological concept that allows the modeling of connections between species in food webs and are thus central to understanding the structure of ecological communities. In this study, a similar model will be fit to the data to determine what parameters are being influenced by temperature, and all for the quantification of interactions strengths between trophic levels.

Known Effects of Warming: Short and Long Term

Predation interactions may be influenced by temperature in many different ways. Here, some short and long term effects of temperature will be discussed. While this study deals primarily with short term effects of warming, it is important to note that rapid temperature changes or short term in vitro experiments may not account for adaptive and evolutionary changes that may occur to offset some of the detrimental effects of warming.

Short Term

There are a few important kinetic effects of warming on predator feeding rates. One, is that warmer temperatures have been shown to significantly increase metabolism, while lowering handling time (most likely due to faster digestion, alongside shorter capture times) (Vucic-Pestic et al. 2011). Furthermore, predator attack rates increased in systems with mobile prey, because faster movement in both organisms meant increased chances for encounters. What was, perhaps, most startling was the decrease in energy efficiency due to increased metabolisms at warmer temperatures. Vucic-Pestic caution that long term effects of warming may include higher susceptibility to starvation due to energy inefficiencies even among abundant prey (2011).

Long Term

The primary long term effect of warming on organisms is a decrease in body size. This was confirmed by a comprehensive study performed by Daufresne et al. in 2009, but had been hypothesized for some time. There are three ecological rules that apply to ecogeographical and ecothermal gradients, which were used as the basis of their hypothesis. The first rule, Bergmann's rule, declares that warm regions tend to have ecological communities dominated by small-sized species (Bergmann 1847). The second rule is the James Rule, which states that within the same species, populations found in warmer environments are smaller (James 1970). Finally, the third rule, the Temperature-Size rule (TSR) follows that as temperatures increase, individual body size of ectotherms decrease (Atkinson 1994). Collectively, these three rules help explain the effects of warming at three important levels: community, population and individual. In a review of published research, along with additional experimental data, Daufresne et al. were able to definitively declare that reduced body size is an effect of global warming on aquatic systems (2009).

Known Effects of Temperature on Functional Responses

The direct effects of body size on functional responses have been measured in the past. Hewett found that capture rates increase with decreasing prey size (1980). In addition, there seem to be contrasting sets of advantages of predators to maintain a large body size (in order to expand its potential range of prey) and to divide as quickly as possible (a process that they found to be most efficient at intermediate, rather than large sizes) (Hewett 1980). Is it possible that decreasing prey sizes help to alleviate or recalibrate the pressure for large body sizes, skewing the trade-off towards more divisions? This assumes that the relative changes in body size are not identical for every species. While this is a little different than my study, these results are useful

in helping me predict how prey size, changing due to temperature increase, may affect functional responses.

One study has looked at the short term effects of temperature on functional response. Ding-xu and Zuo-Rui found that increasing temperatures led to an increased attack rate and decreased handling time in acarophagous thrips preying on hawthorn spider mites, with the functional responses adjusting accordingly (2007).

Research Objectives

The purpose of this study is to illuminate the effects of warming temperatures on ecological systems, via changes in predator-prey dynamics, by pursuing three objectives:

1. Observe potential effects of temperature on population and community dynamics
2. Model the interaction between each trophic level and quantify the magnitude of each interaction strength.
3. Determine the impact of temperature on the magnitude of the trophic cascade.

To reach these objectives, a three-tiered food web was designed and maintained at two different temperatures (17°C and 21°C). Both two-tiered (*Scenedesmus* and *Daphnia*) and three-tiered (*Scenedesmus*, *Daphnia* and *Hydra*) food chains were maintained to assist in isolating shifts in magnitude of the trophic cascade effects. Densities of each population were monitored over a period of 37 days, which provided insights into the population dynamics of each species and the nature and strength of their interactions.

Model System

The model system used included three ubiquitous aquatic genera. The producer was a species of non-motile, colonial algae from the genus *Scenedesmus*. The consumer was *Daphnia*

ambigua, a small planktonic crustacean, ranging from 1-5 millimeters long. The top predator was a species from the genus *Hydra*, a sedentary fresh-water predator.

The genus *Scenedesmus* is very diverse, and one of the most common freshwater genera. The species used was stationary (though some are mobile), and unicellular. However, they have to capacity to form colonies for protection, which has been shown to be common in interactions with *Daphnia* (Hassen and Donk 1993; Lurling 1999). They were selected for use because of their ubiquity, historic relationship with *Daphnia* predation, and ease of rearing.

Daphnia is also a diverse genus that can be found worldwide. They are routinely used as model organisms because of their ubiquity, ease of rearing, fast reproduction, sensitivity to environmental changes, and significance in food webs (Lampert 2006). They are commonly used to study the effects of climate warming on plankton communities and entire ecosystems due to their role as a major link in mediating energy flow between primary producers and secondary consumers in aquatic food webs (Persson et al. 2007). Furthermore, their response to warming temperatures has been researched fairly extensively (Wojtal-Frankiewicz 2012).

Hydra are from the phylum Cnidaria, and have a unique body structure consisting of a tubular body with numerous tentacles attached to an end which are used for stunning and capturing prey. They can be found throughout the fresh waters of North America, and have a high ecological tolerance. (Wright 1997) They were used for this study because of their resilience and ease of rearing. Their long (perhaps indefinite) lifespans and ability to go without food for long periods of time presents a unique challenge while exploring population dynamics. For this reason, *Hydra* populations will be maintained at one individual per microcosm to ensure a steady and reasonable predation pressure across treatments.

Methods

Experimental Biological Community

The *Scenedesmus* was obtained from the lab of Matt Walsh at UT Irvington and was grown in filtered and autoclaved pond water (collected at a freshwater pond on the University of Nebraska-Lincoln's East Campus) at 19°C for three weeks before the experiment. They were grown with light/dark cycles of 12 hours, and fresh pond water was added on every day or every other day. The *Daphnia* were obtained from a pond at Spring Creek Prairie (11700 SW 100th St, Denton, NE 68339) and grown in pond water at 19°C, with 12 hour light/dark cycles. They were fed with *Scenedesmus* every other day, at levels sufficient to maintain population growth. The populations were started 3 weeks before the experiment. The *Hydra* were obtained from Carolina Biological Supply (Burlington, NC, USA), and reared in the 19°C incubator, being fed *Daphnia* for two and a half weeks before the experiment. A group of 12 *Hydra* was maintained in a 150mm-diameter plastic Petri dish, and were fed 20-30 *Daphnia* every other day. The *Hydra* population was starved for two full days before starting the experiment as to ensure uniform hunger for the beginning of the experiment.

Microcosm setup

The biological communities used were set up in lidded, 150mm-diameter plastic Petri dishes. In total, six replicate microcosms were made for each of the four treatments. In each dish (of all treatments), a 90 mm glass rod (with a diameter of 5mm) was glued to the bottom of the dish, along its diameter, with a silicone based glue. Dishes were rinsed three times with deionized water after the glue was applied. The purpose of this rod was to provide potential refuge for *Daphnia*, with the goal of impairing the high predation rate of the *Hydra*, leading to longer population cycling. The rod was not high enough as to completely block organisms from

going over it, and was short enough to allow both *Daphnia* and *Hydra* to go around. To each dish the same amount of water was added, which included 10ml of pond water, 45 mL of algae water (with uniform densities of algae, across treatments), and 3-6 mL of water that accompanied the introduction of the *Daphnia*. Throughout the experiment, to maintain constant water levels, 0.3 mL of pond water was added to each dish to account for evaporation and sampling. On day 0, 10 adult *Daphnia* were placed into each of the two-tiered dishes, and 15 adults and 3 juvenile *Daphnia* were placed into the three-tiered dishes. *Hydra* are very effective predators, so I started with higher *Daphnia* populations to help prolong the population cycle or eventual population extinction. One *Hydra* was placed in each of the three-tiered dishes. To all dishes, 45 mL of the same stock algae/pond water mixture was added.

Temperature Treatments

To observe potential differences in trophic cascade magnitudes between caused by warming temperatures, two temperatures were used that fall within the ranges of survival for all three species used. The two temperatures used were 17°C and 21°C. The microcosms used were placed in two incubators (Percival's Intellus Control System chambers), one with each temperature. All twelve dishes of each temperature treatment were randomly placed on the highest shelf of the corresponding incubator, as to ensure uniform light accessibility, and were rearranged randomly after each measurement.

Data Collection

Population density measurements were collected for each microcosm every two to three days throughout the study period. Populations of all three organisms were measured.
For *Scenedesmus*, densities were determined using a FlowCAM (Fluid Imaging Technologies, Yarmouth, ME, USA). The machine works by running sample liquid over a thin sheet of glass

and taking pictures of each particle that passes by a camera, amplified by a microscope. For each dish, 0.2mL was collected in two 0.1 mL subsamples. Each subsample was pulled from approximately 15mm from the glass rod, and sampled parallel to the rod with the pipette tip being oscillated from the bottom of the dish to the top of the water. Before sampling, each dish was agitated for 10 seconds by gently shaking the dish in horizontal and circular motions. This provided ample agitation to dislodge any algae that had settled to the bottom. To each 0.2 mL sample, 1.8 mL of filtered and autoclaved pond water was added (leading to a 1:10 dilution), and the mixture was vortexed using Digital Vortex Mixer (Fisher Scientific) for approximately 3 seconds. From this mixture, a 1mL portion was pulled and placed into the FlowCAM. The FlowCAM sampled a 0.1ml portion from each dish.

Daphnia densities were manually counted using light tables and the naked eye. Grids of 7 equidistant and parallel lines were applied to a Petri dish lid and placed underneath of each microcosm to aid in accurate counting. Counts were done in random order, and completed twice for each dish with the results averaged. During counting, the dishes were gently shaken to encourage the *Daphnia* to move and to help us differentiate between living non-living organisms. *Hydra* counts were done in the same fashion. Throughout the experiment, *Hydra* populations were maintained at one individual, with any clones present removed during sampling times. This was done to make predation pressure more uniform across repetitions, and to lengthen population cycles.

Data Processing

In order to be used, the raw data from the FLOWCAM required some processing. The machine does not differentiate between live cells and other particulate matter. Each sample needed to be sorted, with non-algal particles removed from the density counts. This process

started by applying a statistical filter, which the program built by determining statistical means and ranges for factors including, but not limited to, length, width, transparency, and circularity, which it determined from a stock of manually selected *Scenedesmus* pictures from the sample list. This filter was very effective in selecting only *Scenedesmus*, but often left out a significant portion of cells. This was in part due to the machine being in and out of focus throughout the duration of the experiment. For each sample, after the filter was applied, the remaining photographs were manually sorted to include all of the *Scenedesmus* present in the sample. Partials were also included. From the refined sample series, the program provided us with algal density per milliliter, taking into account both sample size and dilution.

Mathematical Modeling

To better understand the mechanisms underlying population dynamic shifts and changes in interaction strengths, the data was fit to the Rosenzweig-MacArthur equations (Rosenzweig and MacArthur 1963):

$$\frac{dR}{dt} = rR\left(1 - \frac{R}{K}\right) - f(R)RC \qquad (1)$$

$$\frac{dC}{dt} = ef(R)RC - m(C)C \qquad (2)$$

Here, resource populations grow logistically, with K representing carrying capacity (the level at which the population can reach with zero predation pressure). The potential biomass accumulation present in the resource population when it is in disequilibrium is a function of both K and *r* the maximum growth rate. The fraction of biomass of the consumer lost is represented by *m(*C*).* The functional response of the consumer, *f*(R) is dependent on the biomass of the resource (R), and the attack rate (*a*, not shown). The efficiency of consumption and conversion is

represented by *ef(*R), and relates the amount of biomass gained to the amount of biomass consumed.

After fitting the model to the data using PottersWheel on MATLAB, the parameters were recorded and the appropriate values were plugged into the following equation to quantify interaction strength:

$$\text{Interaction Strength} = \frac{K}{\hat{A}_2} \qquad (1)$$

$$\hat{A}_2 = \frac{d}{a(e - dh)} \qquad (2)$$

Here interaction strength is a function of the carrying capacity of the consumer (K) and the resource density, ($\hat{A}_2$). Death rate of the consumer is represented by *d*, and attack rate by *a*. The Handling time, *h*, is the amount of time the consumer takes to consume a digest the resource before searching again, and *e* is the efficiency of consumption, discussed above.

Results

The results of the study showed complex relationships between temperature, population dynamics, trophic cascades and other potential biological processes. Overall they confirm the understanding of biological communities as complex interactions between species and their environments. The results can be broken down into three segments: (1) effects of temperature on *Daphnia* population dynamics, (2) mathematical modelling results and (3) effects of temperature on trophic cascades.

Overview

The results of the experiment are best explained by system type. In the two-tiered system, *Daphnia* populations saw normal oscillations of boom and bust cycles, typical of *Daphnia* (Figure 1; A). Though significant differences in population dynamics between temperatures were

apparent and will be discussed shortly. *Scenedesmus* demonstrated some response to increasing *Daphnia* populations, with a decline and stabilization of populations (Figure 1; C). In the three-tiered systems, *Daphnia* populations saw sharp declines and eventual extinction by day 15, with *Hydra* populations being maintained at 1 organism per dish (Figure 1; B). There was a slight difference between temperature treatments. *Scenedesmus* showed response to declining *Daphnia* populations, with population increases and stabilizations. In comparing the *Scenedesmus* densities between the two-and three-tiered systems the presence of a strong trophic cascade effect is clear. After the extinction of *Daphnia* in the three-tiered systems, *Scenedesmus* populations spiked and leveled off at (presumably) carrying capacity. This level was almost two orders of magnitude higher than the stable populations of *Scenedesmus* in the two-tiered system.

Effects of Temperature on *Daphnia* Population Dynamics

When the two-tiered microcosm results were analyzed a prominent pattern was observed: that a stark shift in population dynamics occurred between the *Daphnia* populations in the cold and warm temperatures. As Figure 1 demonstrates, the *Daphnia* populations in the warm dishes peaked five days earlier and at a higher population density (an average of 58 versus 50) than the cold populations. In addition, the warm populations went through two boom and bust cycles in the 37 day period as opposed to the cold population's single cycle of growth, decline and stabilization. Lastly, there was more variability in the warm populations in general, with the cold populations showing very similar *Daphnia* densities throughout the study period.

Interestingly, while the *Daphnia* populations showed high variability between temperatures, the *Scenedesmus* populations showed no significant differences. The lack of obvious response to increased predation pressure from higher *Daphnia* populations is worth noting.

In the three-tiered systems, changes in *Daphnia* population dynamics weren't as apparent. We saw a similar trend of increased variability in the warm populations, which makes it hard to provide a clear answer to the differences in population dynamics. Looking at the averages of *Daphnia* population density, though, it appears that warm populations saw increased growth rates (which mirrors the two-tiered results), but were still unable to avoid extinction for any additional length of time. The *Scenedesmus* populations were responsive to the alleviation of predation pressure and rapidly increased with declining *Daphnia* numbers. Their populations were fairly stable for the remainder of the study, with no significant differences between temperature treatments.

Mathematical Modeling

A coupled differential equation model was fit to the data, with relative success (warm: X^2 = 19.1; cold: X^2=56.8). The resulting fit provided the set of parameters found in Table 1, which describe fundamental biological processes. Some parameters were very similar between temperatures including *Daphnia* conservation efficiency and *Daphnia* handling time. Notably, the death rate of *Daphnia* was significantly higher in the warmer populations (1.12 versus 0.27). Furthermore, the attack efficiency for the *Daphnia* was almost twice as high in the warm (0.017 versus 0.10). Carrying capacity was also reported to be an order or magnitude higher in the warm temperature as opposed to the cold ($2.0x10^6$ versus $1.5x10^5$). Using these parameters, interaction strength was quantified using the equation previously discussed. Interaction strength was found to be higher in the warm treatment than in the cold (37.88 versus 1.60).

Effects of Temperature on Trophic Cascades

As previously mentioned, a large trophic cascade effect was present in the three-tiered systems, leading to a steep increase of *Scenedesmus* population density with the decline of

Daphnia. There was no discernable difference in the magnitude of this effect between temperatures, however. To further explore these results, the average density of *Daphnia* was graphed as a function of the density of *Scenedesmus* and time (Figure 2). The blue and red lines, the two-tiered systems, demonstrate circular patterns as is expected with growing and declining population dynamics. The cyan and purple lines demonstrate the three tiered systems (cold and warm, respectively). The shape of these lines is what is important. If the direction and magnitude of these lines differed significantly, a difference in trophic cascade effect would be present. The similar appearances of these two lines support the conclusion that no difference in magnitude was observed.

Discussion

The results lead to two distinct conclusions: (1) *Daphnia* population dynamics are sensitive to temperature variation and (2) the magnitude of the trophic cascade effect showed no temperature dependence. Here, I will discuss these results, and provide potential explanations for my observations. Additionally, I will discuss potential ramifications these results may have on natural systems, areas for improvement, and further research.

Daphnia Population Dynamics

The effect of warmer temperature on the *Daphnia* population dynamics is significant. The warming temperatures acted to create more variability and instability among populations. Where a single cycle and stabilization occurred in the cold, two cycles occurred in the warm over the 37 day period. How might temperature cause this drastic change?

Beisner et al. conducted a similar, two-tiered experiment with *Daphnia* and algae, with similar results. They hypothesized that the primary mechanisms for population destabilization in their experiment was the cumulative direct effects of temperature on biological processes

including birth, death, energy use and rates of feeding (1997). It may also change the timing of population growth in ways that unbalance the predator-prey equilibrium (Murdoch and McCauley 1985).

Murdoch and McCauley went on to hypothesize *Daphnia* population dynamics were heavily influenced by reproductive mechanisms. In cyclical populations, peaks are found when a surge of reproduction creates a cohort of young. Higher densities then work to suppress reproduction, through delayed reproductive development in the young cohort, or suppressed reproduction by adults. Declining populations, often through high death rates in the young cohort, lead to low enough densities to stimulate another reproductive burst, and another cycle begins (Murdoch and McCauley 1987).

The theory that population *Daphnia* population dynamics were highly dependent on birth and death rates was experimentally supported by Pratt in 1943. He found population oscillation at higher temperatures due to stages of high birth rate and low death rate, and stages of little to no birth rate associated with high death rate. He concluded that oscillation consisted of "successive overshooting and under-shooting of a theoretical equilibrium density" which resulted from a delay in the onset of density effects on mortality rates. *Daphnia* populations will remain well over carrying capacity for a few days before completely exhausting their food supply, causing the whole population to collapse. If mortality rates were more closely tied to density, temporally, the population would see less oscillation. (Pratt 1943)

These general theories have been supported by further research looking at how temperature affects specific mechanisms. Reproductive mechanisms are affected by warming temperatures in different ways. First, *Daphnia* have been found to mature faster and reach reproductive maturity more quickly in warmer temperatures (Orcutt and Porter 1984, Hall and

Burns 2002). Weetman found that while reproductive allocation (the amount of energy allocated towards reproduction) depended highly on body weight, temperature significantly affected the slope of the regression so that "maximum reproductive allocation was reached both at an earlier state and smaller size as temperature increased" (2004). This would explain why the warmer *Daphnia* populations peaked sooner in in our experiment. In addition to earlier reproduction, warmer temperature affects brood sizes, though the relationship is slightly more complex. Goss and Bunting found that the largest young production occurred between 15°C and 20°C (1983). Orcutt and Porter found broods were larger at mid-range temperatures (around 15°C) compared to lower temperatures (10°C) (1984). The optimum temperatures they found would actually support larger broods in the colder of our temperature treatments, however. This suggests that the higher peak found in warmer populations may be due more to higher percentages of reproductively active adult *Daphnia* rather than higher brood sizes.

Not only are birth rates affected by temperature, but so is longevity. Orcutt and Porter found that *Daphnia* lived longest at colder temperatures (10°C) and showed declining survivorship with increasing temperatures (1984). This phenomenon shouldn't be surprising, as warming temperatures cause faster maturation and higher metabolic processes, both of which would contribute to shorter life cycles. The effects of temperature aren't quite that simple, though. Orcutt and Porter found that the magnitude of the effects of temperature is dependent on food availability, with temperature dependence being positively correlated with food availability (1984). This may help explain why the second population cycle in the warm populations was smaller in amplitude and longer in length, as reproductive effects of temperature were dampened by lower food availability causing less births and effects on mortality rates were also dampened, leading to lower death rates.

Beisner et al proposed an interesting theory that incorporates many of the aforementioned mechanisms and effects of temperature. They found that in warmer temperatures high rates of juveniles failed to mature. They concluded that higher metabolic requirements due to higher temperatures coupled with low food levels led to a failure to mature, and eventually, death. While small daphnids can survive at lower food densities than mature daphnids, a threshold size exists above which no further growth can occur at low food densities. Beisner observed population dynamics in their two-tiered system and found that *Daphnia* population bursts were caused by pulses of juveniles but declines were caused by a failure of those individuals to mature and reproduce. (1997) This mirrors what was observed in the experimental populations, with high juvenile mortality and little to no full maturation of second generation *Daphnia*.

So far, internal mechanisms of population regulation have been discussed. These are hard to confirm experimentally, since birth and death rates were not recorded, but past research tells us that the changes in these mechanism due to temperature are significant. Of course, this study was set up to focus on predator-prey dynamics, so it is also important to discuss changes in *Daphnia* population dynamics in that context, which can be more directly supported with the data. Namely, the question is: can reactions of *Scenedesmus* populations to changes in *Daphnia* densities, coupled with potential effects of temperature on both species, help to explain the shifts in *Daphnia* population dynamics?

Scenedesmus Responses

Further complicating the interaction strength discussion is the experimental results of the *Scenedesmus* populations and their responses to fluctuating *Daphnia* populations. Inherently, interaction strength is the suppressive effect of one population on another. While methods of quantification vary, one method involves overserving the direct population responses to

predation pressure (i.e. the difference in population density between systems with and without predators). Furthermore, changes in interaction strength may be observed in two structurally identical systems (like the ones used, which only varied by temperature), through observations of relative magnitude (i.e. which system saw large suppressive effects on *Scenedesmus* populations?). If, like the modeling results suggest, interaction strength increased in the warm then intuitively we should see more *Scenedesmus* suppression (lower population levels). Experimentally, that is not the case, as *Scenedesmus* levels do not significantly differ between temperatures. Does that mean interaction strength has remained constant? Not necessarily. It's possible that *Scenedesmus* responses to temperature may be concealing increased interaction strength. Namely, increased temperatures may contribute to increases in productivity. Xin et al. found that *Scenedesmus* expressed logarithmic growth between temperatures of 10 °C and 25 °C, with specific growth rate being positively correlated with increasing temperatures (up to 30°C) (2011). These results were supported by Bouterfas et al in earlier experiments (2002). Higher productivity in the warmer treatments may not have been apparent in the density results because additional growth may have been entirely absorbed by *Daphnia* populations. This additional available energy may have supported increased reproductive allocation in *Daphnia* and contributed to the faster and more variable population dynamics previously discussed.

While this hypothesis may have some validity, it does encounter some potential internal inconsistencies within the experimental results. Following the hypothesis to its full conclusion would mean that we should have seen increased *Scenedesmus* growth in the warm treatments in the three-tiered system after the alleviation of predation pressure due to *Daphnia* extinction. This was not the case, however, as *Scenedesmus* populations did not significantly differ between temperature treatments. Does this definitively mean growth rates were unaffected by

temperature? Not necessarily. It's possible that strong bottom up regulation, whereby *Scenedesmus* growth is limited by resource availability rather than predation pressure, masked any potential differential growth rates. After the alleviation of predation pressure with the *Daphnia* extinction (a release from top-down population regulation), *Scenedesmus* populations quickly reached carrying capacity and may not have been able to express differences in growth rates.

In conclusion, the *Scenedesmus* population results do little to support or detract from the *Daphnia* population dynamic discussion. Further research needs to be done to pinpoint the role temperature plays in the relationships between algal growth rates, *Daphnia* predation, and nutrient availability.

Mathematical Modeling

To further add to the discussion, we can turn to the modeling results, which act to both support aforementioned conclusions and provide a new medium from which to discuss the effects of temperature on interaction strength and trophic cascades.

The mathematical modeling that was used worked to fit the coupled differential equation model to the data. The final results were able to provide values for the parameters needed to quantify the interaction strength between *Daphnia* and *Scenedesmus* populations. Two of these parameters showed significant differences between temperature treatments: attack efficiency (a) and death rate (d). Increased death rate was expected, as found in numerous studies (mentioned above). This strengthens my assertions that *Daphnia* population dynamic shifts occurred, in part, due to longevity effects. The other parameter, increased attack efficiency, was also to be expected. Englund at al.'s meta-analysis found that, across species, there is a positive correlation between higher attack efficiency and increasing temperatures, up to a point (24.9 C +- 2.5 C)

(2011). This trend was also supported by Vucic-Pestic et al., whose model supported increasing attack efficiency due to temperature (2011). This effect may be due to faster kinetic movement in *Daphnia*, and may be magnified by the stationary nature of the *Scenedesmus* , because similar kinetic effects on prey mobility would potentially act to offset the same effect on *Daphnia* individuals.

The significance of the modeling results is most profound in the conclusion that interaction strength between the *Daphnia* and *Scenedesmus* populations increased at the warmer temperature. Interestingly, the significantly higher death rate of *Daphnia* (more than five times higher) alone would suggest a decreased interaction strength, while higher attack efficiency (though less than twice as high) would support increased interaction strength. The reason the high death rate did not lead to calculations of a lower interaction strength, was because the fitted models also provided estimates of carrying capacity that were significantly different. The models suggest that the warm temperatures could support *Scenedesmus* populations an order of magnitude higher (10^6 as opposed to 10^5) than colder temperatures. This large positive difference outweighed the negative effect of increased death rates in determining the magnitude and direction of the shift in interaction strength. While some literature has shown increased potential carrying capacity due to warming (Xin et al 2011), it was not supported by the experimental results. Under the three-tiered system, with the extinction of *Daphnia*, *Scenedesmus* populations of both temperature treatments oscillated at around the same density ($1X10^6$). Because no predation pressure was present, this number presumably represents the realized carrying capacity of *Scenedesmus* in the system, and showed no temperature dependence. This contradiction between modeling and experimental results may point to weaknesses in the modeling results, and

therefore make it hard to decisively say that interaction strength was indeed different between the two temperatures.

Trophic Cascade Effects

The unique purpose of this study was to determine the effects of temperature on the magnitude of trophic cascades. The results provide a clear answer: that the magnitude of the trophic cascade in question, the indirect affect the *Hydra* had on *Scenedesmus* due to the release of *Daphnia* predation pressure, demonstrated no significant change between the two temperature treatments.

It's important to note that an observable change in the magnitude trophic cascade would be difficult to isolate, if one were present. Ultimately, for algae populations to be affected by a trophic cascade, there would need to be differential top-down regulatory pressure on the *Daphnia*. This brings up two issues.

First, in this experiment, this differential in regulatory pressure could only have come from changes in *Hydra* predation efficiency or differential reproductive responses due to the presence of *Hydra* (though, this may be more unlikely). As it turns out, this experimental design may have been too limiting to observe a trophic cascade effect because it eliminated an important variable: differential population dynamics of *Hydra*. While this set up was intentional, it ended up relying on the single variable of *Hydra* attack efficiency to determine changes in the trophic cascade. While the goal was to simplify the experiment, it ended up complicating a question that could have been resolved with a single *Hydra-Daphnia* system, where attack efficiency could have been directly observed. Furthermore, by using a sedentary top predator, I

effectively minimized the chances of observing changes in trophic cascade due to temperature because, as Novich et al. found, sedentary predators have temperature-independent interaction strengths (2014).

Unfortunately, the data was not robust enough to model the predator-prey relationship between *Hydra* and *Daphnia* which may have provided insights into potential changes in attack efficiency or interaction strength as a whole. Observationally, it is possible to infer that attack efficiency may have increased, because the *Hydra* in the warm treatments drove (on average) higher populations of *Daphnia* extinct after the apparent but brief *Daphnia* population burst between days five and ten. There is no way to confirm the validity of this observation, however, as it is likely strongly affected by the high level of *Daphnia* population variability in the warmer temperature.

Second, biological systems are highly complex, even when simplified as much as this. While the experiment was set up in such a way (with both two- and three- tiered systems) as to allow differentiation of temperature dependent effects of *Daphnia* populations on *Scenedesmus* populations, and apply that to the three-tiered system as a means of isolating the effects of *Hydra*, it's hard to say observational differences in *Scenedesmus* densities could definitively be tied to the indirect effects of *Hydra*. As previously discussed, it is hard to completely explain the population dynamics of *Scenedesmus*, and especially so since there are no stand-alone results demonstrating temperature dependent growth.

In summary, the results indicate that the trophic cascade present in this system did not respond to changes in temperature. It should be noted, however, that the experiment allowed for only a limited potential effect, due to the maintenance of constant top-predation pressure. Therefore, these results may have little application to natural biological systems.

Implications

Changes in Population and Community Dynamics

This research adds to the understanding that population dynamics are sensitive to changing temperatures. This could have serious ramifications as climactic changes continue to accelerate. In fact, some researchers have already documents effects. One general consequence of warming temperatures, increased variability in population dynamics, when coupled with increased metabolic requirements may work to increase the rate of extinction among a wide range of species (Vuvic-Pestic at al. 2011). It is important to note that the relationship between temperature and performance is highly variable between species (Goldman 1977). This is caused differences in thermal sensitivity and by differences in temperature minimums, maximums and optimal ranges for any given biological function (Kordas 2011).

It is because of these temperature response differences that some of the most significant changes will occur, changes to community dynamics and structure. Wojtal-Frankiewicz, in her review of *Daphnia* population dynamics concluded that temperature may play a large role in affecting community dynamics and shifting species dominance (2012). In one study by Winder and Schindler, warming spring temperatures have led to a mismatch in the onset of algal growth and *Daphnia* population cycles which led to a significant decrease in *Daphnia* populations and a shift in species abundance to a *Daphnia* species that was less temperature sensitive (2007). However, a similar study at a different location by Huber et al. found no such decoupling response (2008). Wagner and Bennforf. went as far to say that their data suggest that "even low warming by 1.7°C during a short, but critical seasonal period, resulting in the coincidence of two or more factors adversely affecting a keystone species, such as *Daphnia*, may induce changes in whole lake food webs and thus alter entire ecosystems" (2007)

While we know that there is a large potential for significant effects of warming, these effects will be highly variable and situational. Huber et al. found that temperature dependent effects on a natural food web that included numerous species of algae, *Daphnia* and other zooplankton, were highly dependent on nutrient availability. They concluded that "an assessment of possible changes in nutrient loading is crucial when anticipating how phytoplankton could evolve under future climate warming" (2008). This highlights the importance of considering the complexity of natural systems when predicting responses to warming.

Changes in Trophic Cascade

In regards to trophic cascades, not only is temperature dependency little understood, but also the ramifications of any potential effects have yet to be fully explored. What is known is that the magnitudes of trophic cascades are highly variable, but they can strongly shape community dynamics in many systems. Potential shifts in magnitude may have serious restructuring or destabilizing effects on systems, worldwide.

Plasticity and Evolution

Another compounding factor in this discussion is the role plasticity and evolution will play in supporting species resilience (and consequently, community resilience) to warming temperatures. Little is known about the long term effects of warming temperatures on predator-prey relationships and community dynamics. It is possible that some short term effects that contribute to instability (like reproductive mechanisms) may be counteracted by evolutionary adaptation.

<u>Areas for Improvement</u>

To improve the strength of this study and allow a more precise look at the temperature dependence of trophic cascades I would recommend the following changes:

1. Alter biological system
2. Enhance experimental design
3. Collect more robust data

Alter Biological System

One of the complicating factors, as discussed above, was the inability to observe changes in *Hydra* population dynamics. The culling of *Hydra* was a fix to a broken system. It avoided the problem of ever-expanding populations and led to slightly longer *Daphnia* cycles. The reality is that *Hydra* was not the best choice to study trophic cascades because it does not express typical population dynamics. Choosing a top predator who reflected typical population dynamics would allow for a more robust and realistic understanding of the temperature dependence of trophic cascades.

Enhance Experimental Design

In order to better understand the complexity inherent in three-tiered food chains, and better isolate potential changes in trophic cascades, more experiments should be run addressing more of the individual components of the system. Culturing isolated *Scenedesmus* populations at both temperatures would have provided a definitive answer regarding the temperature dependence of its growth in this system. Maintaining the *Scenedesmus-Daphnia* system is still important, but it should be complimented by *Daphnia-Hydra* systems as well. This would allow for the direct observation of both individual interaction strengths inherent in the system, and would allow for a more accurate understanding of the three-tiered system results.

Increasing both the variability and number of temperature treatments may also help in observing potential effects, and would add to the applicability of the study. The temperatures used were arguably both within the optimum range of all three species. Additionally, I would consider adding nutrients to the system in order to decrease bottom-up regulation and observe more directly the effects of predation.

Collect More Robust Data

While I was limited by time and resources for this particular study, more repetitions could have been useful by reducing some of the noise and variation among populations (especially in the warm treatment). Furthermore, more specific observations including population age demographics, reproductive events, and mortality would add much needed clarity to the explanation of relevant mechanisms.

Further Areas of Research

This study and associated literature review highlights the need for continued research on the temperature dependence of trophic cascades, as well as other mechanisms involved in shaping population and community dynamics. While observational studies in natural environments have alluded to potential disruptive effects of warming on biological communities, more in vitro research may be helpful in clarifying and understanding important mechanisms that may be muddled in vivo. More research could be done to understand more confounding variables, like the temperature dependence of defense responses or predator and prey behavior.

Conclusion

This study was well equipped to observe changes in community dynamics due to temperature. It found that *Daphnia* population dynamics are highly temperature dependent, and

hypothesized underlying temperature-dependent mechanisms. These included rates of maturation and mortality, reproductive timing and capacity, and *Scenedesmus* productivity. The experimental design allowed for a limited trophic cascade response to temperature, and saw no effects. The experiment would need expanding to better explore trophic cascade effects. What was aiming to be an exploration of trophic cascades may have only effectively added to the discussion of population dynamic and direct predator-prey interactions. Regardless, the observed population dynamic shifts, which echo many previous studies, should add to concerns about the potential detrimental and destabilizing effects of global climate change on biological communities.

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References

Atkinson, D. "Temperature and organism size: a biological law for ectotherms?." *Advances in ecological research* 25 (1994): 1-58.

Barbosa, Pedro A., ed. *Conservation biological control*. Academic Press, 1998.

Beisner, Beatrix E., Edward McCauley, and Frederick J. Wrona. "The influence of temperature and food chain length on plankton predator prey dynamics." Canadian Journal of Fisheries and Aquatic Sciences 54.3 (1997): 586-595.

Bergmann, C. "About the relationships between heat conservation and body size of animals." *Goett Stud* 1 (1847): 595-708.

Bouterfas, Radia, Mouhssine Belkoura, and Alain Dauta. "Light and temperature effects on the growth rate of three freshwater [2pt] algae isolated from a eutrophic lake." Hydrobiologia 489.1-3 (2002): 207-217.

Carpenter, Stephen R., James F. Kitchell, and James R. Hodgson. "Cascading trophic interactions and lake productivity." *BioScience* (1985): 634-639.

Clarke, B. R. Y. A. N. "Balanced polymorphism and the diversity of sympatric species." *Taxonomy and geography* 4 (1962): 47-70.

Daufresne, Martin, Kathrin Lengfellner, and Ulrich Sommer. "Global warming benefits the small in aquatic ecosystems." *Proceedings of the National Academy of Sciences* 106.31 (2009): 12788-12793.

Ding-Xu, Li, Tian Juan, and Shen Zuo-Rui. "Functional response of the predator Scolothrips takahashii to hawthorn spider mite, Tetranychus viennensis: effect of age and temperature." *BioControl* 52.1 (2007): 41-61.

Englund, Göran, et al. "Temperature dependence of the functional response." Ecology letters 14.9 (2011): 914-921.

Glasser, John W. "The role of predation in shaping and maintaining the structure of communities." *American Naturalist* (1979): 631-641.

Goldman, Joel C. "Temperature effects on phytoplankton growth in continuous culture." Limnology and Oceanography 22.5 (1977): 932-936.

Goss, L. Barry, and Dewey L. Bunting. "*Daphnia* development and reproduction: responses to temperature." Journal of Thermal Biology 8.4 (1983): 375-380.

Hall, Catherine J., and Carolyn W. Burns. "Mortality and growth responses of *Daphnia* carinata to increases in temperature and salinity." Freshwater Biology 47.3 (2002): 451-458.

Hessen, Dag O., and Ellen Van Donk. "Morpholigical changes in *Scenedesmus* induced by substances released from *Daphnia*." *Archiv für Hydrobiologie* 127 (1993).

Hewett, Steve W. "The effect of prey size on the functional and numerical responses of a protozoan predator to its prey." *Ecology* 61.5 (1980): 1075-1081.

Huffaker, Carl B., ed. *Theory and practice of biological control*. Elsevier, 2012.

James, Frances C. "Geographic size variation in birds and its relationship to climate." *Ecology* 51.3 (1970): 365-390.

Karl, Thomas R., et al. "Global warming: Evidence for asymmetric diurnal temperature change." *Geophysical Research Letters* 18.12 (1991): 2253-2256.

Knight, Tiffany M., et al. "Trophic cascades across ecosystems." *Nature*437.7060 (2005): 880-883.

Lampert, Winfried. "*Daphnia*: model herbivore, predator and prey." *Polish journal of ecology* 54.4 (2006): 607-620.

Lürling, Miquel. *The smell of water: grazer-induced colony formation in Scenedesmus*. Landbouwuniversiteit Wageningen, 1999.

McCauley, Edward, and William W. Murdoch. "Cyclic and stable populations: plankton as paradigm." American Naturalist (1987): 97-121.

Murdoch, William W., and Edward McCauley. "Three distinct types of dynamic behaviour shown by a single planktonic system." (1985): 628-630.

Novich, Rachel A., et al. "The temperature independence of interaction strength in a sit-and-wait predator." Ecosphere 5.10 (2014): art137.

Orcutt Jr, John D., and Karen G. Porter. "The synergistic effects of temperature and food concentration of life history parameters of *Daphnia*." Oecologia 63.3 (1984): 300-306.

Paine, Robert T. "Food web complexity and species diversity." *American Naturalist* (1966): 65-75.

Persson, Jonas, et al. "Food quantity and quality regulation of trophic transfer between primary producers and a keystone grazer (*Daphnia*) in pelagic freshwater food webs." *Oikos* 116.7 (2007): 1152-1163.

Peterson, A. Townsend, Monica Papes, and Daniel A. Kluza. "Predicting the potential invasive distributions of four alien plant species in North America." (2009).

Pratt, David M. "Analysis of population development in *Daphnia* at different temperatures." The Biological Bulletin 85.2 (1943): 116-140.

Real, Leslie A. "The kinetics of functional response." *American Naturalist*(1977): 289-300.

Ripple, William J., and Robert L. Beschta. "Wolves and the ecology of fear: can predation risk structure ecosystems?." *BioScience* 54.8 (2004): 755-766.

Rosenzweig, Michael L., and Robert H. MacArthur. "Graphical representation and stability conditions of predator-prey interactions." *American Naturalist* (1963): 209-223.

Schalau, Kristine, et al. "Temperature is the key factor explaining interannual variability of *Daphnia* development in spring: a modelling study." Oecologia 157.3 (2008): 531-543.

Shurin, Jonathan B., et al. "A cross-ecosystem comparison of the strength of trophic cascades." *Ecology letters* 5.6 (2002): 785-791.

Vallina, Sergio M., and Corinne Le Quéré. "Stability of complex food webs: Resilience, resistance and the average interaction strength." *Journal of Theoretical Biology* 272.1 (2011): 160-173.

VUCIC-PESTIC, O. L. I. V. E. R. A., et al. "Warming up the system: higher predator feeding rates but lower energetic efficiencies." Global Change Biology 17.3 (2011): 1301-1310.

Wagner, Annekatrin, and Jürgen Benndorf. "Climate-driven warming during spring destabilises a *Daphnia* population: a mechanistic food web approach." Oecologia 151.2 (2007): 351-364.

Walther, Gian-Reto, et al. "Ecological responses to recent climate change." *Nature* 416.6879 (2002): 389-395.

Weetman, D., and D. Atkinson. "Evaluation of alternative hyes to explain temperature-induced life history shifts in *Daphnia*." Journal of Plankton Research 26.2 (2004): 107-116

Winder, Monika, and Daniel E. Schindler. "Climate change uncouples trophic interactions in an aquatic ecosystem." *Ecology* 85.8 (2004): 2100-2106.

Wojtal-Frankiewicz, Adrianna. "The effects of global warming on *Daphnia* spp. population dynamics: a review." Aquatic Ecology 46.1 (2012): 37-53

Wootton, J. Timothy, and Mark Emmerson. "Measurement of interaction strength in nature." *Annual Review of Ecology, Evolution, and Systematics* (2005): 419-444.

Wright, Johnathan. "*Hydra*." *Hydra*. Northern State University, 1 Jan. 1997. Web. 21 Apr. 2015.

Xin, Li, Hu Hong-Ying, and Zhang Yu-Ping. "Growth and lipid accumulation properties of a freshwater microalga *Scenedesmus* sp. under different cultivation temperature." Bioresource technology 102.3 (2011): 3098-3102.

Figure 1. Graphs of the algae and *Daphnia* densities, across time, for both temperatures and both two and three-tiered systems.

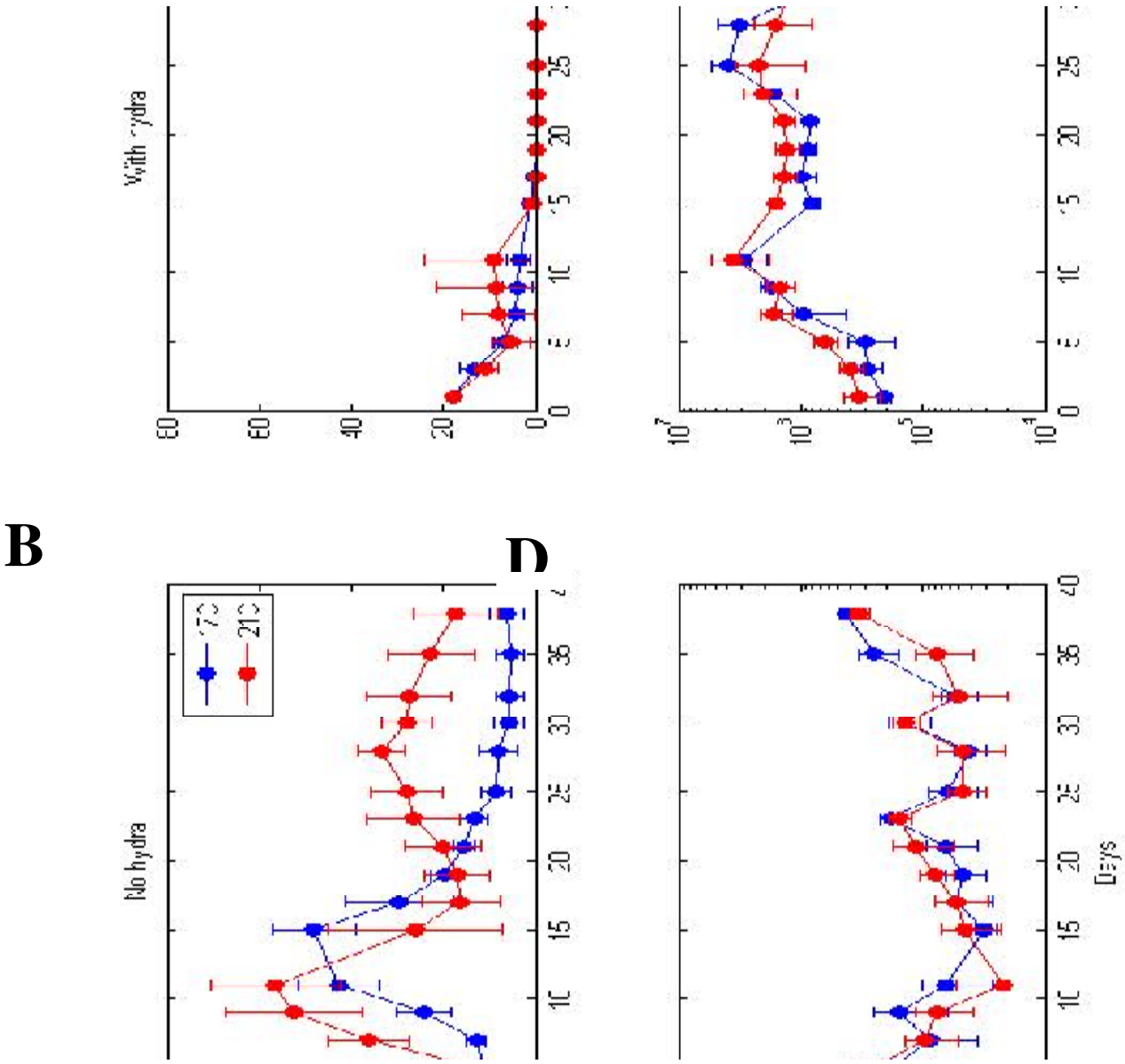

Figure 2. Graphs plotting *Daphnia* density against algae density, for both temperatures (E is 17°C, F is 21°C), and for the three-tiered (cyan and magenta) and two-tiered (blue and red).

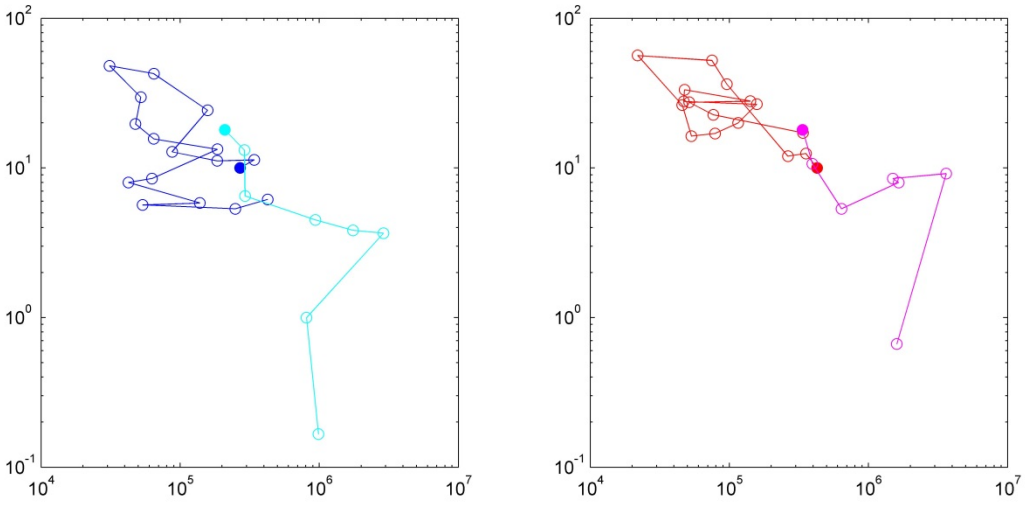

Figure 3. Data with best model fits, which were used to determine parameters necessary for the quantification of interaction strength.

Warm (21°C) Cold (17°C)

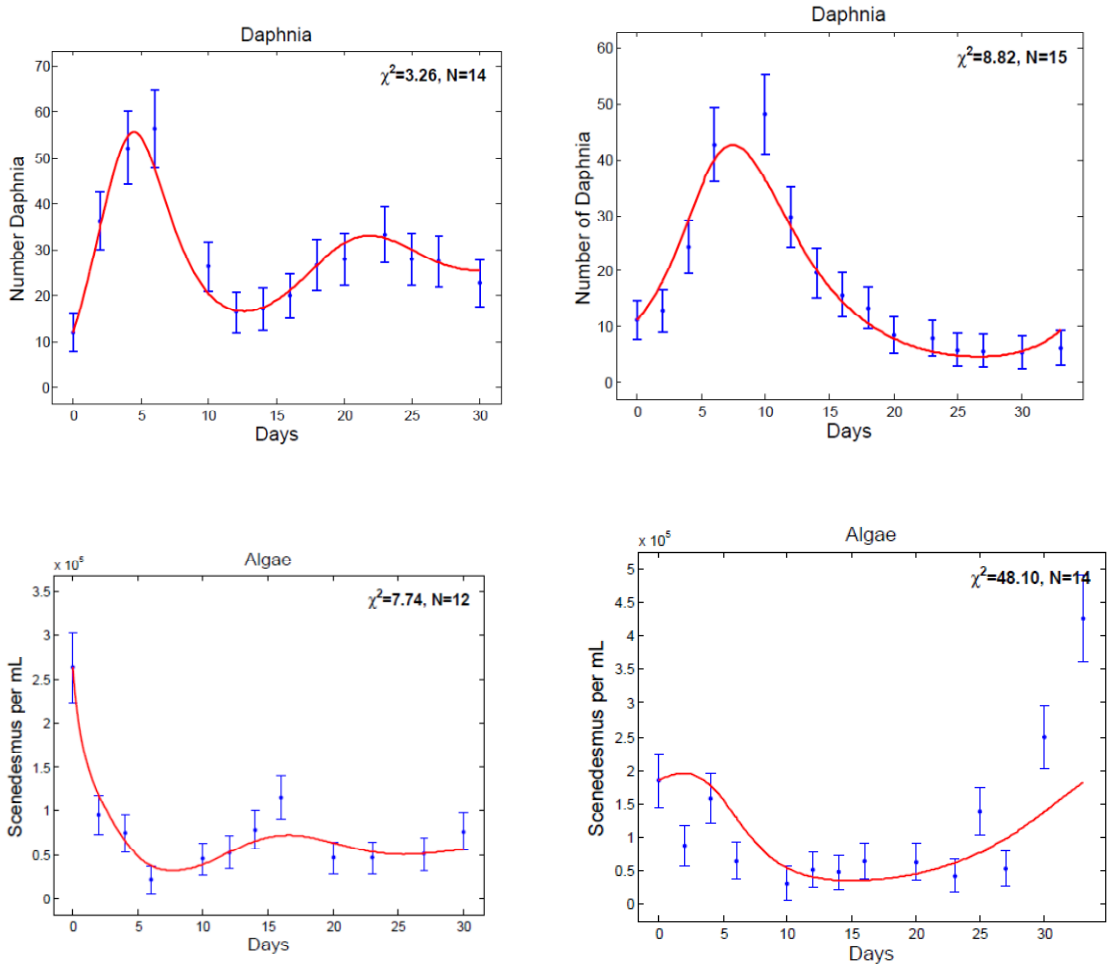

Table 1. Parameters pulled from the model fittings which were used to quantify interaction strength.

Parameters	**Description**	**Cold (17° C)**	**Warm (17° C)**

d	Death rate (*Daphnia*)	0.27	1.12
a	Attack efficiency (*Daphnia*)	0.01	0.017
e	Conversion efficiency (*Daphnia*)	0.00031	0.00023
h	Handling time	0.000082	0.00094
k	Carrying capacity	$1.5X10^5$	$2.0X10^6$
	Interaction Strength	1.60	37.88